species which are immobilized in said gene transcript pattern probe kit and assessing the amount of hybridization, so as to determine the degree of correlation indicative of the presence of said disease or condition or stage thereof, and so as to diagnose or identify said disease or condition or a stage thereof in said test [prokaryotic or] eukaryotic organism.

492

REMARKS

In paragraph 1, on page 2 of the Office Action, the Examiner alleges that the application fails to meet the sequence listing requirement because a computer readable form (CRF) was not filed.

Contrary to the Examiner's contention, a Sequence Listing, along with a CRF, was filed on February 1, 2000 (a copy of which is attached hereto, along with the filing receipt therefor).

The Examiner also requests that Applicants amend pages 29-31 of the specification to refer to the sequence ID numbers.

Accordingly, Applicants hereby amend the specification as requested by the Examiner.

In paragraph 2, on page 2 of the Office Action, the Examiner notes that the present application is a Continuation of PCT/GB98/01261 under 35 U.S.C. § 120, not a §371 Application, as indicated on the Official Filing Receipt. The Examiner suggests that Applicants may wish to request a Corrected Official Filing Receipt to correct this error.

Accordingly, file simultaneously herewith a Request for Corrected Official Filing Receipt to correct the above-noted error.

In paragraph 5, on page 2 of the Office Action, the Examiner rejects Claim 31 under 35 U.S.C. § 112, second paragraph.

Specifically, the Examiner states that Claim 31 is indefinite because it depends on a later claim.

Accordingly, Applicants hereby amend Claim 31 to include the recitations of Claim 33 therein, thereby rendering moot the Examiner's rejection.

In paragraph 7, on page 3 of the Office Action, the Examiner rejects Claims 18-25 and 27 under 35 U.S.C. § 102(b) as being anticipated by Wadhwa et al.

Specifically, the Examiner states that Wadhwa et al teaches a method of isolating selected cDNA species comprising:

(a) isolating mRNA from a normal mouse cell line, reverse transcribing the mRNA, amplifying the cDNA and labeling the resulting cDNA with denaturing loading dye;

(b) isolating mRNA from a transformed clone, reverse transcribing the mRNA, amplifying the cDNA and labeling the resulting cDNA with denaturing loading dye;

(c) separating the cDNA species using gel electrophoresis;

(d) selecting two or more cDNA species from the separated cDNA species obtained in step (c),

which are present at a different level in the normal sample than in the diseased sample;

(e) isolating and amplifying the resulting selected cDNA species; and

(f) immobilizing the resulting isolated selected cDNA species on a membrane filter.

For the following reasons, Applicants respectfully traverse the Examiner's rejection.

The Examiner is requested to note that Claim 18 has been amended to refer to the isolation of mRNA from a sample obtained from a part of an organism distant to the area of said disease. This amendment is supported in the specification, at page 10, last paragraph, and on page 23, line 15 *et seq*. The Examiner is also requested to note that the Examples in the present application perform the method of the invention in this manner. In line with this amendment, steps (a) and (b) have been reversed, and thus revised step (b) now refers to isolating material from a site corresponding to the site of the source of the tissue, cells or body fluid for the "diseased sample". Further, the organisms have been limited to "eukaryotic" organisms, since the amended claims are not applicable to unicellular organisms, such as some prokaryotes.

Finally, Claim 18 has been amended to refer to isolating mRNA from one or more diseased organisms. Thus, the development is of a disease standard specific to the disease, and is not related to a specific individual's gene expression. This amendment is supported by the specification at page 4, lines 14-17, page 5, first paragraph and page 21, lines 23-29.

For the following reasons, Claim 18 as amended, and its dependent claims are clearly novel over Wadhwa et al.

Wadhwa et al teaches a reverse Northern analysis to identify gene transcripts which are differentially expressed in a cell line and its transformed derivative.

Wadhwa et al deals with transformed cell lines, and thus is not concerned with isolating probes characteristic of a disease or condition or stage thereof as required in amended Claim 18. In addition, the sample which is investigated in Wadhwa et al is not derived from a part of the organism that is distant to the area of said disease, as required by amended Claim 18. Instead in Wadhwa et al, the nucleic acid molecules are derived directly from the "diseased", i.e., transformed, tissue.

Accordingly, Applicants respectfully submit that Wadhwa et al does not teach or suggest the present invention, and thus request withdrawal of the Examiner's rejection.

In paragraph 8, on page 4 of the Office Action, the Examiner rejects Claims 18, 21-23, 25-26 and 28 under 35 U.S.C. § 102(b) as being anticipated by Graver et al.

Specifically, the Examiner states that Graver et al teaches a method of obtaining isolated selected cDNA species which comprises:

(a) isolating mRNA from a normal esophageal mucosa tissue sample, reverse transcribing the mRNA, and amplifying the cDNA;

(b) isolating mRNA from a carcinoma of the esophagus sample, reverse transcribing the mRNA and amplifying the cDNA;

(c) separating the cDNA species using gel electrophoresis;

(d) selecting two or more cDNA species from the separated cDNA species obtained in step (c) which are present at a different level in the normal sample than in the diseased sample; and

(e) isolating the resulting selected cDNA species by excision from the gel.

For the following reasons, Applicants respectfully traverse the Examiner's rejection.

Graber et al teaches the identification of differentially expressed genes in normal relative to diseased tissues. As mentioned in connection with Wadhwa et al, the sample which is investigated in Graber et al is derived directly from diseased tissue, and thus falls outside amended Claim 18.

Accordingly, Applicants respectfully submit that Graver et al does not teach or suggest the present invention, and thus request withdrawal of the Examiner's rejection.

Although no obviousness rejections have been raised specifically against the above-noted claims in light of Wadhwa et al and Graber et al, the Examiner is requested to note that in both references, genes which are differentially expressed in normal versus transformed/diseased tissue are examined. In contrast, in the present invention, the transformed/diseased tissue is <u>not</u> examined. Instead, samples are tested which are not primarily affected by disease, e.g., cells found in the blood (e.g., as exemplified in the Examples) or other body fluid, which reflect changes which are indicative of a disorder present

elsewhere in the organism. Thus, for example, whereas the diseased tissue might show an increase in expression of various oncogenic factors, these factors might not be expressed in cells from samples taken elsewhere from the body. The cells in these samples are thus, likely to show altered expression of quite different genes, e.g., genes related to metabolism.

Hence, the probes which are isolated in the present invention which can be used to characterize a particular disease are an entirely different set to those which might be isolated from the disease site. The identification of probes from non-disease tissue is not taught or suggested in any of the prior art in which, in all cases, differential expression in diseased relative to normal tissue is examined. There was also no motivation provided to do so and there is no recognition that differential expression might be observed in distant tissues, cells or body fluid, as claimed in the present invention. The present invention provides an alternative non-invasive method of isolating suitable probes and their use in diagnosis which are unobvious over the prior art.

In paragraph 9, on page 5 of the Office Action, the Examiner rejects Claims 29-31 under 35 U.S.C. § 102(e) as being anticipated by Pinkel et al.

Specifically, the Examiner states that Pinkel et al teaches kits which comprise a solid support having an array of target nucleic acids bound thereto, and a container containing nucleic acids representing the normal reference genome or cDNA from a reference cell type (see column 3, lines 41-49 thereof). Further, the Examiner states that Pinkel et al teaches that the

nucleic acids on the array may be cDNA or RNA (see column 2, lines 50-54).

For the following reasons, Applicants respectfully traverse the Examiner's rejection.

The kits of Pinkel et al are not suitable for the claimed purpose, i.e., they are not suitable for diagnosing or identifying a disease or condition or stage thereof. This is because the technique of Pinkel et al does not derive probes which correspond to those isolated by the claimed method.

The probes of the present invention are different from those isolated from diseased tissue. Thus, unless one started with the same samples, regardless of how differentially expressed genes were identified, the same genes (and thus, probes related to those genes) can not be identified. It should be noted in this respect that Pinkel et al deals with probes isolated from normal or diseased tissue (see column 3, lines 15 *et seq*). Nowhere is it indicated that cells, tissue or body fluid distant to the disease site should be used to generate probes, as claimed in the present invention. As a consequence, any probes isolated in accordance with Pinkel et al would be distinct to those referred to in Claim 29.

Furthermore, Claim 29 requires that the solid support carries the probe species. In contrast, Pinkel et al binds the target genetic material to a solid support, which is then probed to identify binding of that probe to the immobilized target material. In the present invention, the probe is immobilized on the solid support, and the target genetic material is bound to the immobilized probe.

As mentioned above with respect to Wadhwa et al and Graber et al, since Pinkel et al does not recognize that probes isolated from tissue, cells or body fluid distant to the site of disease may be informative for the preparation of a kit for diagnosis of disease, there is no motivation in Pinkel et al to seek such probes or make kits containing them.

Accordingly, Applicants respectfully submit that Pinkel et al does not teach or suggest the present invention, and thus request withdrawal of the Examiner's rejection.

In paragraph 11, on page 6 of the Office Action, the Examiner rejects Claims 29-34 under 35 U.S.C. § 103 as being obvious over Wadhwa et al in view of the Stratagene Catalog.

Specifically, the Examiner notes that Wadhwa et al does not teach packaging of the immobilized cDNA species thereof into a kit, nor a method for making a kit. However, the Examiner states that the Stratagene Catalog teaches gene characterization kits, and thus one of ordinary skill in the art would have been motivated to use the method disclosed by Wadhwa et al to produce a kit containing the cDNAs on a solid support and other reagents useful for gene transcript comparisons, such as a normal and transformed sample, as taught by Wadhwa et al to be used in nucleic acid research, since the Stratagene Catalog teaches the benefits of kits.

For the following reasons, Applicants respectfully traverse the Examiner's rejection.

The Stratagene Catalog simply identifies the value of kits (in this case, an unrelated kit) in providing prepackaged reagents and quality control for particular assays.

As mentioned above, however, Wadhwa et al does not teach or suggest the identification of probes which correspond to the probes referred to in Claim 29. Without the motivation to identify such probes, there would be no motivation to produce a kit, and thus such kits are similarly unobvious. Methods using such unobvious kits must similarly be viewed as being unobvious.

The Examiner is requested to note that Claim 32 has been amended in line with Claim 18. Thus, it is directed to methods of preparing kits, and directly refers to methods of isolating the appropriate probes in a manner which is quite distinct to the method used to isolate the probes in Wadhwa et al, as mentioned above with respect to Claim 18. Similarly, methods of preparing transcript patterns using the claimed kits require that the target mRNA (or cDNA) which is examined is isolated from a sample from a site distant to the disease site. Since both the kits are unobvious (as discussed above) and the target molecules which are examined are not suggested or even in keeping with the methods described in Wadhwa et al, Claims 33 and 34 are similarly unobvious over the cited references.

Accordingly, Applicants respectfully submit that the present invention is not taught or suggested in Wadhwa et al, alone or in view of the Stratagene Catalog, and thus request withdrawal of the Examiner's rejection.

Finally, in paragraph 12, on page 7 of the Office Action, the Examiner rejects Claim 35 under 35 U.S.C. § 103 as being obvious over Wadhwa et al in view of the Stratagene Catalog, and in further view of Seilhamer et al.

Specifically, the Examiner states that Wadhwa et al in view of the Stratagene Catalog do not teach a method in which a test sample is compared to a known sample for diagnosis of disease. However, the Examiner states that Seilhamer et al teaches that gene transcripts from a biological specimen can be quantified and compared against the transcripts of a diseased and healthy patient in order to diagnose a disease (see page 12, lines 5-20 thereof).

Thus, the Examiner contends that it would have been *prima facie* obvious to one of ordinary skill in the art to include in such a comparison step in the method taught by Wadhwa et al in order to provide a method for diagnosis of disease, since Seilhamer et al teaches that such comparisons are useful for disease diagnosis.

For the following reasons, Applicants respectfully traverse the Examiner's rejection.

Claim 35 is directed to a method of diagnosis using the claimed assay by the generation of hybridization patterns.

Seilhamer et al teaches isolation of transcripts from diseased patients and comparison to reference databases of normal and diseased samples. The Examiner refers to the passage at page 12, lines 5-20 of Seilhamer et al which illustrates that transcripts from diseased and healthy patients may be compared. It will however be noted, see page 12, lines 20-23 of Seilhamer et al, that the transcripts of normal cells or tissues are compared to those <u>from diseased cells or tissues</u>. Thus, in contrast to the present invention, there is <u>no</u> indication in Seilhamer et al that the tissue, cells or body fluids distant to

the site of disease might be used for the comparison. Hence, Seilhamer et al does not overcome the deficiencies in Wadhwa et al and the Stratagene Catalog to provide a motivation to use nucleic acid material isolated from cells, tissue or body fluids distant to the site of disease. Claim 35 specifically states the source of the genetic material (see steps (a) and (c)), and thus must be viewed as unobvious over the cited references.

Accordingly, Applicants respectfully submit that the present invention is not taught or suggested in Wadhwa et al, alone or in view of the Stratagene Catalog and Seilhamer et al, and thus request withdrawal of the Examiner's rejection.

In view of the amendments to the specification and claims and the arguments set forth above, reexamination, reconsideration and allowance are respectfully requested.

The Examiner is invited to contact the undersigned at his Washington telephone number on any questions which might arise.

Respectfully submitted,

Gordon Kit
Registration No. 30,764

SUGHRUE, MION, ZINN,
MACPEAK & SEAS, PLLC
2100 Pennsylvania Avenue, N.W.
Washington, D.C. 20037-3202
(202) 293-7060

Date: October 20, 2000